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NEWS HOURS	STN Operating Hours Plus Help Desk Availability
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FILE 'HOME' ENTERED AT 14:01:10 ON 06 JUL 2004

=> file ca biosis medline

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FILE 'BIOSIS' ENTERED AT 14:01:27 ON 06 JUL 2004
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FILE 'MEDLINE' ENTERED AT 14:01:27 ON 06 JUL 2004

=>  s lysosmotropic?
L1          13 LYSOSMOTROPIC?

=> s lysosomotropic?
L2        3080 LYSOSOMOTROPIC?

=> s chloroquine
L3       29635 CHLOROQUINE

=> s hydroxychloroquine
L4        3034 HYDROXYCHLOROQUINE

=> s primaquine
L5        4017 PRIMAQUINE

=> s methylamine
L6       31444 METHYLAMINE

=> s l1 or l2 or l3 or l4 or l5 or l6
L7       66767 L1 OR L2 OR L3 OR L4 OR L5 OR L6

=> s antibody or mab or mcab or moab
L8     1638482 ANTIBODY OR MAB OR MCAB OR MOAB

=> s (cell or plasma)(W)(surface or membrane)
   2 FILES SEARCHED...
L9      706438 (CELL OR PLASMA)(W)(SURFACE OR MEMBRANE)

=> s l9 or cd?
L10    1971481 L9 OR CD?

=> s l7 and l8 and l10
L11        732 L7 AND L8 AND L10

=> s fluoresc? or fluorophore or stain? or label? or FACS or (flow(W) cytometr?)
L12    2541190 FLUORESC? OR FLUOROPHORE OR STAIN? OR LABEL? OR FACS OR (FLOW(W)
                CYTOMETR?)

=> s l11 and l12
L13        251 L11 AND L12

=> l10(10a)l12
L10(10A)L12 IS NOT A RECOGNIZED COMMAND
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"HELP COMMANDS" at an arrow prompt (=>).

=> s l10(10a)l12
L14      79360 L10(10A) L12

=> s l7 and l8 and l14
L15         49 L7 AND L8 AND L14

=> d his

     (FILE 'HOME' ENTERED AT 14:01:10 ON 06 JUL 2004)
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     FILE 'CA, BIOSIS, MEDLINE' ENTERED AT 14:01:27 ON 06 JUL 2004
L1           13 S LYSOSMOTROPIC?
L2         3080 S LYSOSOMOTROPIC?
L3        29635 S CHLOROQUINE
L4         3034 S HYDROXYCHLOROQUINE
L5         4017 S PRIMAQUINE
L6        31444 S METHYLAMINE
L7        66767 S L1 OR L2 OR L3 OR L4 OR L5 OR L6
L8      1638482 S ANTIBODY OR MAB OR MCAB OR MOAB
L9       706438 S (CELL OR PLASMA)(W)(SURFACE OR MEMBRANE)
L10     1971481 S L9 OR CD?
L11         732 S L7 AND L8 AND L10
L12     2541190 S FLUORESC? OR FLUOROPHORE OR STAIN? OR LABEL? OR FACS OR (FLOW
L13         251 S L11 AND L12
L14       79360 S L10(10A)L12
L15          49 S L7 AND L8 AND L14

=> dupe rem
DUPE IS NOT A RECOGNIZED COMMAND
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"HELP COMMANDS" at an arrow prompt (=>).

=> duplicate remove
ENTER L# LIST OR (END):l13
DUPLICATE PREFERENCE IS 'CA, BIOSIS, MEDLINE'
KEEP DUPLICATES FROM MORE THAN ONE FILE?  Y/(N):n
PROCESSING COMPLETED FOR L13
L16          155 DUPLICATE REMOVE L13 (96 DUPLICATES REMOVED)

=> duplicate remove
ENTER L# LIST OR (END):l15
DUPLICATE PREFERENCE IS 'CA, BIOSIS, MEDLINE'
KEEP DUPLICATES FROM MORE THAN ONE FILE?  Y/(N):n
PROCESSING COMPLETED FOR L15
L17           24 DUPLICATE REMOVE L15 (25 DUPLICATES REMOVED)

=> d l17 1-24 bib ab

L17  ANSWER 1 OF 24  CA  COPYRIGHT 2004 ACS on STN     DUPLICATE 1
AN   140:139305  CA
TI   Early microglial activation following neonatal excitotoxic brain damage in
     mice: a potential target for neuroprotection
AU   Dommergues, M.-A.; Plaisant, F.; Verney, C.; Gressens, P.
CS   Service de Neurologie Pediatrique, Hopital Robert-Debre, INSERM E 9935,
     Paris, 75019, Fr.
SO   Neuroscience (Oxford, United Kingdom) (2003), 121(3), 619-628
     CODEN: NRSCDN; ISSN: 0306-4522
PB   Elsevier Science Ltd.
DT   Journal
LA   English
AB   Previous studies in a mouse model of neonatal excitotoxic brain damage
     mimicking the brain lesions in human cerebral palsy showed microglial
     activation within 24 h after intracerebral injection of the glutamatergic
     analog ibotenate.  Using this model, we studied the expression of CD-45
     antigen, a marker of blood-derived cells, by these activated microglial
     cells labeled by Griffonia simplicifolia I isolectin B4.  Immunohistochem.
     performed during early development of excitotoxic lesions showed that most
     cells labeled with the isolectin B4 were CD-45-neg.,
     suggesting that these early activated microglial cells were deriving
     chiefly from resident microglia and not from circulating monocytes.  We
     also directly tested the hypothesis that activated resident microglia
```

post dates

and/or blood-derived monocytes play a role in the pathophysiol. of excitotoxic brain damage. Repeated i.p. administrations of **chloroquine, chloroquine** + colchicine, minocycline, or an anti-MAC1 **antibody** coupled to the toxin saporin before and/or after ibotenate injection induced a significant reduction in the d. of isolectin B4-pos. cells. This inhibition of resident microglial and/or blood-derived monocytes activation was accompanied by a significant reduction in the severity of ibotenate-induced brain lesions (up to 79% lesion size reduction with the highest minocycline dose) as well as of ibotenate-induced cortical caspase-3 activation (49% reduction).

RE.CNT 30 THERE ARE 30 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L17 ANSWER 2 OF 24 CA COPYRIGHT 2004 ACS on STN DUPLICATE 2

AN 139:20758 CA

TI Flow cytometry based detection of HLA alloantibody mediated classical complement activation

AU Wahrmann, Markus; Exner, Markus; Regele, Heinz; Derfler, Kurt; Kormoczi, Gunther F.; Lhotta, Karl; Zlabinger, Gerhard J.; Bohmig, Georg A.

CS Department of Internal Medicine III, Division of Nephrology and Dialysis, University of Vienna, Vienna, A-1090, Austria

SO Journal of Immunological Methods (2003), 275(1-2), 149-160
CODEN: JIMMBG; ISSN: 0022-1759

PB Elsevier Science B.V.

DT Journal

LA English

AB Complement-dependent cytotoxicity (CDC) panel reactive **antibody** (PRA) testing is used to assess recipient presensitization and post-transplant alloantibody formation in transplant recipients. However, CDC test results can be affected by false-pos. reactions brought about by autoantibodies or antilymphocyte reagents. As an alternative to the CDC-PRA assay, detection of HLA alloantibodies using HLA antigen-coated microbeads (FlowPRA test) was recently established. FlowPRA testing, however, does not distinguish between (presumably more harmful) complement-fixing and noncomplement-fixing alloantibodies. Here, the authors established a novel assay allowing flow cytometric detection of HLA alloantibody dependent classical complement activation using the FlowPRA test. For the detection of complement activation, FlowPRA beads were incubated with sera from highly sensitized dialysis patients (**CDC**-PRA reactivity >60%) and then **stained** for C4 (C4d, C4c) and C3 (C3d, C3c) fragments, as well as C1q deposition using indirect immunofluorescence. The authors demonstrate alloantibody induced induction of C4 fragment, and in parallel C1q deposition to HLA class I or class II beads. As shown by immunoblotting, C4 staining was not due to the presence of preformed C4 fragment-IgG/IgM complexes. Indeed, C4 fragment deposition in this in vitro system was demonstrated to result from de novo complement activation. First, inactivation of C4 by treatment of sera with **methylamine**, which inhibits cleavage of the internal thioester, completely abolished C4 fragment deposition. Second, C4 fragment deposition was not observed in the evaluation of C4-free immunoadsorption eluates obtained from highly sensitized dialysis patients. After supplementation with complement, however, eluates induced C4 deposition. Deposition of C4 split products and C1q was temperature-dependent with maximum binding after incubation at 4° for 60 min. In contrast, maximum C3 fragment deposition was found at 37°. At this temperature, C3 deposition occurred in an alloantibody and C4-independent fashion, presumably as a result of alternative complement activation. Thus, the authors describe a novel cell-independent and easy-to-perform PRA test that permits flow cytometry based detection of alloantibody induced classical complement activation.

RE.CNT 41 THERE ARE 41 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L17 ANSWER 3 OF 24 CA COPYRIGHT 2004 ACS on STN
AN 137:108283 CA
TI Quantitation of HLA-DR and CD11b expression on peripheral blood cells
IN Davis, Kenneth A.
PA Becton, Dickinson and Company, USA
SO U.S., 23 pp., Cont.-in-part of U.S. 6,200,766.
CODEN: USXXAM
DT Patent
LA English
FAN.CNT 2

instant pat family

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	US 6423505	B1	20020723	US 1999-406013	19990924
	US 6200766	B1	20010313	US 1998-204860	19981203
	WO 2000033082	A2	20000608	WO 1999-US28884	19991202
	WO 2000033082	A3	20001130		
	W: JP, US				
	RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
	EP 1135685	A2	20010926	EP 1999-964122	19991202
	R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
	US 6376202	B1	20020423	US 2000-645966	20000824
	US 2002076734	A1	20020620	US 2002-80868	20020222
PRAI	US 1998-204860	A2	19981203		
	US 1999-406013	A2	19990924		
	WO 1999-US28884	W	19991202		

AB The author discloses an improved method and reagents for quantitation of HLA-DR and/or CD11b expression on peripheral blood cells. The method comprises inclusion of a **lysosomotropic** amine, such as **chloroquine**, during **staining** which stabilizes HLA-DR and **CD11b** surface expression. Use of an anti-CD14 conjugate, anti-CD14-PerCP/CY5.5, permits the ready discrimination of monocytes. The improved method and reagents may be used to assess immune competence, and to direct and monitor immunostimulatory therapies in immune suppression associated with sepsis.

RE.CNT 68 THERE ARE 68 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L17 ANSWER 4 OF 24 CA COPYRIGHT 2004 ACS on STN DUPLICATE 3
AN 137:134784 CA
TI **Hydroxychloroquine** reverses platelet activation induced by human IgG antiphospholipid **antibodies**

post dates

AU Espinola, Ricardo G.; Pierangeli, Silvia S.; Ghara, Azzudin E.; Harris, E. Nigel
CS Department of Microbiology, Biochemistry and Immunology, Atlanta, GA, USA
SO Thrombosis and Haemostasis (2002), 87(3), 518-522
CODEN: THHADQ; ISSN: 0340-6245
PB Schattauer GmbH
DT Journal
LA English
AB Prothrombotic properties of antiphospholipid (aPL) **antibodies** may be explained in part by their ability to enhance the activation of platelets pre-treated with low doses of ADP or thrombin. The antimalarial drug **hydroxychloroquine** (HQ) has been used successfully in prevention of postoperative thrombosis and in treatment of patients with SLE or APS. In one study, administration of HQ reversed the thrombogenic properties of aPL in mice. However, the mechanism of action of HQ in preventing thrombosis is not clearly understood. In order to explore this further, the effects of HQ on activation of platelets by aPL in the presence of a thrombin agonist was studied. The changes in the expression of GPIIb/IIIa (**CD41a**) and GPIIIa (**CD61**) on platelet membrane by **flow cytometry** were used as indicators of

platelet activation. Citrated whole blood from a healthy donor was treated at room temperature with suboptimal doses of a thrombin agonist receptor peptide (TRAP) and affinity-purified aPL **antibodies**, in the presence and in the absence of **hydroxychloroquine** (1 mM). TRAP increased the expression of GPIIb/IIIa and GPIIIa on platelet surface. The treatment of the platelets with the six aPL **antibodies** in the presence of 12 nmol/mL TRAP further increased the expression of GPIIb/IIIa by 42.3±12.3% and the expression of GPIIIa was further incremented by 46.8±13.5%. The effects of aPL and TRAP on expression of platelet surface markers of activation was completely abrogated by HQ in a dose-dependent fashion and was effective at concns. of HQ as low as 25 μg/mL (0.0125 mM). This suggests at least one possible mechanism by which HQ may prevent thrombosis. This may have important implications in treatment of thrombosis in APS patients.

RE.CNT 32 THERE ARE 32 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L17 ANSWER 5 OF 24 CA COPYRIGHT 2004 ACS on STN DUPLICATE 4
AN 136:52478 CA
TI Membrane trafficking of CD1c on activated T cells
AU Salamone, Maria Del C.; Mendiguren, Ana Karina; Salamone, Gabriela V.; Fainboim, Leonardo
CS Immunogenetics Division, University Hospital, School of Medicine, University of Buenos Aires, Argent.
SO Journal of Leukocyte Biology (2001), 70(4), 567-577
CODEN: JLBIE7; ISSN: 0741-5400
PB Federation of American Societies for Experimental Biology
DT Journal
LA English
AB The authors investigated the regulation of and the intracellular trafficking involved in the membrane expression of CD1c antigen on activated mature T cells. Membrane expression of this glycoprotein was highly regulated and dependent on the activation state of the cells. The presence of the CD1c antigen on activated peripheral blood mononuclear cells (PBMCs) was confirmed by flow cytometry, reverse transcriptase-PCR (RT-PCR), and immunoperoxidase staining. The RT-PCR anal. of the α3- and 3'-untranslated regions of CD1C showed that phytohemagglutinin (PHA) activation induced expression of transcripts that encode the three isoforms (soluble, membrane, and cytoplasmic/soluble). Immunocytochem. studies showed a specific association of CD1c with the cell membrane and a cytoplasmic, perinuclear distribution. Although **flow-cytometric staining** confirmed the intracellular presence of **CD1c**, membrane expression on PHA blast cells was not detected. The authors found that membrane detection of CD1c antigen was temperature dependent. Cell surface binding of the anti-CD1c monoclonal **antibody** (**mAb**) was consistently neg. at 4 and 37° but was detected at room temperature (18-22°). At physiol. temps., activated PBMCs showed intracellular accumulation of the anti-CD1c **mAbs**, indicating that CD1c cycled between cell surface and intracellular compartments. The CD1c exocytosis pathway was sensitive to Brefeldin A, cytochalasin B, and **chloroquine**.

RE.CNT 54 THERE ARE 54 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L17 ANSWER 6 OF 24 MEDLINE on STN
AN 2001360164 MEDLINE
DN PubMed ID: 11423908
TI Evidence of a lysosomal pathway for apoptosis induced by the synthetic retinoid CD437 in human leukemia HL-60 cells.
AU Zang Y; Beard R L; Chandraratna R A; Kang J X
CS Department of Medicine, Massachusetts General Hospital and Harvard Medical School, Boston, MA 02114, USA.

NC CA-79553 (NCI)
SO Cell death and differentiation, (2001 May) 8 (5) 477-85.
Journal code: 9437445. ISSN: 1350-9047.
CY England: United Kingdom
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200108
ED Entered STN: 20010813
Last Updated on STN: 20010813
Entered Medline: 20010809
AB The novel synthetic retinoid 6-[3-(1-adamantyl)-4-hydroxyphenyl]-2-naphtalene carboxylic acid (AHPN/CD437) has been proven to be a potent inducer of apoptosis in a variety of tumor cell types. However, the mechanism of its action remains to be elucidated. Recent studies suggest that the lysosomal protease cathepsin D, when released from lysosomes to the cytosol, can initiate apoptosis. In this study, we examined whether cathepsin D and free radicals are involved in the CD437-induced apoptosis. Exposure of human leukemia HL-60 cells to CD437 resulted in rapid induction of apoptosis as indicated by caspase activation, phosphatidylserine exposure, mitochondrial alterations and morphological changes. Addition of the antioxidants alpha-tocopherol acetate effectively inhibited the CD437-induced apoptosis. Measurement of the intracellular free radicals indicated a rise in oxidative stress in CD437-treated cells, which could be attenuated by alpha-tocopherol acetate. Interestingly, pretreatment of cells with the cathepsin D inhibitor pepstatin A blocked the CD437-induced free radical formation and apoptotic effects, suggesting the involvement of cathepsin D. However, Western blotting revealed no difference in cellular quantity of any forms of cathepsin D between control cells and CD437-treated cells, whereas immunofluorescence analysis of the intracellular distribution of cathepsin D showed release of the enzyme from lysosomes to the cytosol. **Labeling** of lysosomes with **lysosomotropic** probes confirmed that **CD437** could induce lysosomal leakage. The CD437-induced relocation of cathepsin D could not be prevented by alpha-tocopherol acetate, suggesting that the lysosomal leakage precedes free radical formation. Furthermore, a retinoic acid nuclear receptor (RAR) antagonist failed to block these effects of CD437, suggesting that the action of CD437 is RAR-independent. Taken together, these data suggest a novel lysosomal pathway for CD437-induced apoptosis, in which lysosomes are the primary target and cathepsin D and free radicals act as death mediators.

L17 ANSWER 7 OF 24 MEDLINE on STN
AN 2001261532 MEDLINE
DN PubMed ID: 11315636
TI Involvement of vacuolar proton ATPase in Junin virus multiplication.
AU Castilla V; Palermo L M; Coto C E
CS Laboratorio de Virologia, Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires, Buenos Aires, Argentina.
SO Archives of virology, (2001) 146 (2) 251-63.
Journal code: 7506870. ISSN: 0304-8608.
CY Austria
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 200105
ED Entered STN: 20010521
Last Updated on STN: 20010521
Entered Medline: 20010517
AB The role of vacuolar-proton ATPase (V-H+ ATPAse) on Junin virus (JV) replication was evaluated by analyzing the effect of specific inhibitors of the enzyme activity on different steps of virus multiplication cycle.

The presence of the macrolide antibiotics bafilomycin A1 and concanamycin A during the first two hours of infection caused a significant reduction of extracellular infectious virus production and viral protein expression in Vero and BHK-21 cells. The inhibitory action of the compounds was mainly exerted at an early stage of the JV multiplication cycle, without affecting virus attachment to the cell but preventing virus penetration. A correlation between the inhibitory action of the compounds on intracellular compartments acidification and the reduction of JV yield was observed. The addition of concanamycin A at different times after infection indicated that the compound also interferes with the release of infectious particles to the extracellular medium. Although, intracellular transport of JV glycoproteins to the **cell membrane**, seems not to be affected as revealed by immunofluorescence **staining**. The results confirm that JV enters into the cell through the endocytic pathway as previously suggested by using **lysosomotropic** compounds.

L17 ANSWER 8 OF 24 CA COPYRIGHT 2004 ACS on STN
AN 133:16300 CA
TI Improved methods and reagents for quantitation of HLA-DR and CD11b expression on peripheral blood cells
IN Davis, Kenneth A.
PA Becton Dickinson and Company, USA
SO PCT Int. Appl., 60 pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 2

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	WO 2000033082	A2	20000608	WO 1999-US28884	19991202
	WO 2000033082	A3	20001130		
	W: JP, US				
	RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
	US 6200766	B1	20010313	US 1998-204860	19981203
	US 6423505	B1	20020723	US 1999-406013	19990924
	EP 1135685	A2	20010926	EP 1999-964122	19991202
	R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, FI				
PRAI	US 1998-204860	A2	19981203		
	US 1999-406013	A2	19990924		
	WO 1999-US28884	W	19991202		

AB Improved methods, reagents, and kits for quantitation of HLA-DR and/or CD11b expression on peripheral blood cells are presented. Inclusion of a **lysosomotropic** amine, such as **chloroquine**, during **staining** stabilizes HLA-DR and **CD11b** expression. Use of a novel anti-CD14 conjugate, anti-CD14-PerCP/CY5.5, permits the ready discrimination of monocytes. The improved methods, reagents, and kits can be used to assess immune competence, and to direct and monitor immunostimulatory therapies in patients exhibiting monocyte deactivation.

L17 ANSWER 9 OF 24 CA COPYRIGHT 2004 ACS on STN DUPLICATE 5
AN 134:110215 CA
TI Rhodamine 123 binds to multiple sites in the multidrug resistance protein (MRP1)
AU Daoud, Roni; Kast, Christina; Gros, Philippe; Georges, Elias
CS Institute of Parasitology, Macdonald Campus, Ste-Anne-de-Bellevue, Can.
SO Biochemistry (2000), 39(50), 15344-15352
CODEN: BICHAW; ISSN: 0006-2960
PB American Chemical Society
DT Journal
LA English

AB The mechanisms of MRP1-drug binding and transport are not clear. In this study, we have characterized the interaction between MRP1 and rhodamine 123 (Rh123) using the photoreactive-iodinated analog, [125I]iodoaryl azido-rhodamine 123 (or IAARh123). Photoaffinity **labeling** of **plasma membranes** from HeLa cells transfected with MRP1 **cDNA** (HeLa-MRP1) with IAARh123 shows the photolabeling of a 190 kDa polypeptide not labeled in HeLa cells transfected with the vector alone. Immunopptn. of a 190 kDa photolabeled protein with MRP1-specific monoclonal **antibodies** (QCRL-1, MRPr1, and MRPm6) confirmed the identity of this protein as MRP1. Anal. of MRP1-IAARh123 interactions showed that photolabeling of membranes from HeLa-MRP1 with increasing concns. of IAARh123 was saturable, and was inhibited with excess of IAARh123. Furthermore, the photoaffinity labeling of MRP1 with IAARh123 was greatly reduced in the presence of excess Leukotriene C4 or MK571, but to a lesser extent with excess doxorubicin, colchicine or **chloroquine**. Cell growth assays showed 5-fold and 14-fold increase in the IC50 of HeLa-MRP1 to Rh123 and the Etoposide VP16 relative to HeLa cells, resp. Anal. of Rh123 fluorescence in HeLa and HeLa-MRP1 cells with or without ATP suggests that cross-resistance to Rh123 is in part due to reduced drug accumulation in the cytosol of HeLa-MRP1 cells. Mild digestion of purified IAARh123-photolabeled MRP1 with trypsin showed two large polypeptides (.apprx.111 and .apprx.85 kDa) resulting from cleavage in the linker domain (L1) connecting the multiple-spanning domains MSD0 and MSD1 to MSD2. Exhaustive proteolysis of purified IAARh123-labeled 85 and 111 kDa polypeptides revealed one (6 kDa) and two (.apprx.6 plus 4 kDa) photolabeled peptides, resp. Resolution of total tryptic digest of IAARh123-labeled MRP1 by HPLC showed three radiolabeled peaks consistent with the three Staphylococcus aureus V8 cleaved peptides from the Cleveland maps. Together, the results of this study show direct binding of IAARh123 to three sites that localize to the N- and C-domains of MRP1. Moreover, IAARh123 provides a sensitive and specific probe to study MRP1-drug interactions.

NR

RE.CNT 54 THERE ARE 54 CITED REFERENCES AVAILABLE FOR THIS RECORD
ALL CITATIONS AVAILABLE IN THE RE FORMAT

L17 ANSWER 10 OF 24 MEDLINE on STN
AN 97402254 MEDLINE
DN PubMed ID: 9259318
TI Intracellular trafficking of angiotensin II and its AT1 and AT2 receptors: evidence for selective sorting of receptor and ligand.
AU Hein L; Meinel L; Pratt R E; Dzau V J; Kobilka B K
CS Falk Cardiovascular Research Center and Department of Medicine, Stanford University School of Medicine, California 94305, USA.
SO Molecular endocrinology (Baltimore, Md.), (1997 Aug) 11 (9) 1266-77.
Journal code: 8801431. ISSN: 0888-8809.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199710
ED Entered STN: 19971021
Last Updated on STN: 19980206
Entered Medline: 19971006

NR

AB Angiotensin II (Ang II) binds to two different receptor subtypes, AT1 and AT2 receptors. In many cases, receptor stimulation by Ang II is followed by a rapid desensitization of the intracellular signal transduction and a decrease in cell surface receptor number. The present study was designed to examine by immunofluorescence microscopy the cellular trafficking pathways of Ang II and its AT1a and AT2 receptors in human embryonal kidney 293 cells stably expressing these receptor subtypes. Fluorescently labeled Ang II and AT1a receptors were rapidly internalized into endosomes. AT2 receptors were localized in the plasma membrane and did not undergo endocytosis upon agonist stimulation. After removal of

agonist, AT1a receptors recycled to the **plasma membrane**, whereas **fluorescently labeled** Ang II was targeted to the lysosomal pathway. Even though no further loss of surface receptor was measurable by ligand binding at steady state, fluorescein-Ang II was continuously internalized, and cycling of receptor between endosomal vesicles and the plasma membrane was detected by **antibody** feeding. These experiments provide evidence for subtype-specific receptor sorting and internalization of Ang II and its AT1a receptor as a receptor-ligand complex, and they suggest that the sequestration of receptors into endosomes is in dynamic equilibrium with receptor cycling to the plasma membrane. Finally, internalization of AT1a receptors and Ang II persists after desensitization mechanisms have attenuated Ca2+ and inositol 1,4,5-trisphosphate signaling.

L17 ANSWER 11 OF 24 CA COPYRIGHT 2004 ACS on STN DUPLICATE 6
AN 127:13196 CA
TI Induction of apoptosis in peripheral blood lymphocytes following treatment in vitro with **hydroxychloroquine**
AU Meng, Xue Wei; Feller, John M.; Ziegler, John B.; Pittman, Sally M.; Ireland, Christine M.
CS Sydney Children's Hospital, Sydney, NSW 2031, Australia
SO Arthritis & Rheumatism (1997), 40(5), 927-935
CODEN: ARHEAW; ISSN: 0004-3591
PB Lippincott-Raven
DT Journal
LA English
AB Defective regulation of apoptosis may be central to the development of autoimmune disorders. This study investigated the possibility that the antirheumatic effect of hydroxycholoroquine (HCQ) may be achieved by up-regulation of apoptosis. Peripheral blood lymphocytes collected from normal controls and patients with systemic lupus erythematosus (SLE) were cultured in the presence or absence of a range of concns. of HCQ. Cells undergoing apoptosis were identified by several standard methods, including morphol. changes, DNA fragmentation, and flow cytometry. For some expts., lymphocytes were simultaneously **stained** with **antibodies** to T **cell surface** markers and with propidium iodide for dual-**stain flow cytometric** studies. HCQ was able to induce apoptosis in peripheral blood lymphocytes in a dose- and time-dependent manner. HCQ induced these changes in all T cell subpopulations studied. There was no significant difference between the controls and patients with SLE in terms of the percentage of apoptotic cells detected following treatment with HCQ. The present study demonstrated that HCQ induces apoptosis in peripheral blood lymphocytes, which leads to the speculation that HCQ may exert its antirheumatic effect through this mechanism.

L17 ANSWER 12 OF 24 MEDLINE on STN
AN 95031956 MEDLINE
DN PubMed ID: 7524481
TI Agonist-induced internalization of the substance P (NK1) receptor expressed in epithelial cells.
AU Garland A M; Grady E F; Payan D G; Vigna S R; Bunnett N W
CS Department of Surgery, University of California, San Francisco.
NC DK 39957 (NIDDK)
DK 43207 (NIDDK)
NS 21710 (NINDS)
SO Biochemical journal, (1994 Oct 1) 303 (Pt 1) 177-86.
Journal code: 2984726R. ISSN: 0264-6021.
CY ENGLAND: United Kingdom
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199411

ED Entered STN: 19941222
Last Updated on STN: 19970203
Entered Medline: 19941109

AB Internalization of the NK1 receptor (NK1R) and substance P was observed in cells transfected with cDNA encoding the rat NK1R by using anti-receptor **antibodies** and cyanine 3-labelled substance P (cy3-substance P). After incubation at 4 degrees C, NK1R immunoreactivity and cy3-substance P were confined to the plasma membrane. Within 3 min of incubation at 37 degrees C, NK1R immunoreactivity and cy3-substance P were internalized into small intracellular vesicles located beneath the plasma membrane. Fluorescein isothiocyanate-labelled transferrin and cy3-substance P were internalized into the same vesicles, identifying them as early endosomes. After 60 min at 37 degrees C, NK1R immunoreactivity was detected in larger, perinuclear vesicles. Internalization of 125I-labelled substance P was studied by using an acid wash to dissociate **cell-surface label** from that which has been internalized. Binding reached equilibrium after incubation for 60 min at 4 degrees C with no detectable internalization. After 10 min incubation at 37 degrees C, 83.5 +/- 1.0% of specifically bound counts were internalized. Hyperosmolar sucrose and phenylarsine oxide, which are inhibitors of endocytosis, prevented internalization of 125I-labelled substance P and accumulation of NK1R immunoreactivity into endosomes. Acidotropic agents caused retention of 125I-labelled substance P within the cell and inhibited degradation of the internalized peptide. Continuous incubation of cells with substance P at 37 degrees C reduced 125I-substance P binding at the cell surface. Therefore, substance P and its receptor are internalized into early endosomes within minutes of binding, and internalized substance P is degraded. Internalization depletes NK1Rs from the cell surface and may down-regulate the response of a cell to substance P.

NR

L17 ANSWER 13 OF 24 CA COPYRIGHT 2004 ACS on STN DUPLICATE 7
AN 120:70218 CA
TI Ligand-dependent polyubiquitination of c-kit gene product: a possible mechanism of receptor down modulation in M07e cells
AU Miyazawa, Keisuke; Toyama, Keisuke; Gotoh, Akihiko; Hendrie, Paul C.; Mantel, Charlie; Broxmeyer, Hal E.
CS 1st Dep. Intern. Med., Tokyo Med. Coll., Tokyo, Japan
SO Blood (1994), 83(1), 137-45
CODEN: BLOOAW; ISSN: 0006-4971
DT Journal
LA English

checked full article 7/6/04 DKJ

AB Quantities of proteins in cells are balanced by protein synthesis and degradation Protein ubiquitination is an important ATP-dependent proteolytic pathway for "short-lived" proteins. The authors show that soluble steel factor (SLF) stimulation at 37° rapidly induced polyubiquitination of c-kit protein in growth-factor-dependent human-myeloid cell line M07e, resulting in smeared, retarded migration of c-kit protein in SDS-PAGE in the mol. weight region of 145 kDa. Receptor ubiquitination was almost completely absent when cells were treated with SLF at 4° or at 37° in the presence of 0.2% sodium azide, or when the cells were pretreated with anti-c-kit monoclonal **antibody** or genistein, a tyrosine kinase inhibitor. This suggested that c-kit ubiquitination was ligand dependent and appeared to require intrinsic tyrosine-kinase activation of the c-kit protein. **Flow-cytometric** anal. of c-kit expression on the **cell surface** of M07e cells showed down modulation of c-kit within 5 min after soluble-SLF treatment at 37°. However, rapid receptor down modulation was almost completely suppressed when cells were treated with SLF at 4° or at 37° in the presence of 0.2% sodium azide, conditions that concomitantly suppressed polyubiquitination of c-kit protein. In addition, these conditions almost completely suppressed radiolabeled SLF (125I-SLF) internalization after ligand-receptor interaction. Pulse-chase studies of

35S-methionine-labeled c-kit protein showed that SLF stimulation at 37° strikingly enhanced c-kit degradation (T1/2; .apprx.20 min) compared with that in cells stimulated with SLF at 4° or at 37° with 0.2% sodium azide. However, in the presence of **chloroquine**, which blocks lysosomal degradation, this ligand-induced c-kit degradation at 37° was only suppressed in part. These data suggest that SLF-induced polyubiquitination of the c-kit receptor protein may play a role in regulation of c-kit-encoded protein-receptor expression in M07e cells.

L17 ANSWER 14 OF 24 CA COPYRIGHT 2004 ACS on STN DUPLICATE 8
AN 120:52177 CA
TI Major histocompatibility complex class I-binding peptides are cycled to the cell surface after internalization
AU Abdel Motal, Ussama M.; Zhou, Xianzheng; Joki, Annalena; Siddiqi, Abdur Rehman; Srinivasa, B. R.; Stenvall, Kristina; Dahmen, Jan; Jondal, Mikael
CS Dep. Immunol., Karolinska Inst., Stockholm, S-171 77, Swed.
SO European Journal of Immunology (1993), 23(12), 3224-9
CODEN: EJIMAF; ISSN: 0014-2980
DT Journal
LA English
AB Cytotoxic T lymphocytes (CTL) recognize target antigens as short, processed peptides bound to major histocompatibility complexes class I (MHC-I) heavy and light chains (β2-microglobulin; β2-m). The heavy chain, which comprise the actual peptide binding α-1 and α-2 domains, can exist at the cell surface in different forms, either free, bound to β2-m or as a ternary complex with β2-m and peptides. MHC-I chains are also known to internalize, and recycle to the cell surface, and this has been suggested to be important in peptide presentation. Whether MHC-I-bound peptides also can recycle is not known. The authors have investigated this by using both peptide transporter mutant RMA-S cells and EL4 cells loaded with Db-binding peptides, by two different approaches. First, peptides were covalently linked with galabiose (Galα4Gal) at a position which did not interfere with Db binding or immunogenicity, and peptide recycling tested with Gal2-specific monoclonal **antibodies**. By **flow cytometry**, a return of Gal2 epitopes to the **cell surface** as found, after cellular internalization and cell surface clearance by pronase treatment. This peptide recycling could be discriminated from free fluid-phase uptake and was inhibited by **methylamine**, **chloroquine** and low temperature (18°) but not by leupeptin. Second, specific CTL were reacted with peptide-loaded target cells after complete removal of surface Db mols. by pronase, and after different times of incubation at 37° to allow reexpression. By this procedure, reappearance of target cell susceptibility was confirmed. The results are in agreement with a model for optimizing peptide presentation by recycling through an intracellular compartment similar to early endosomes in certain antigen-presenting cells.

L17 ANSWER 15 OF 24 CA COPYRIGHT 2004 ACS on STN DUPLICATE 9
AN 117:46249 CA
TI Spontaneous and ligand-induced endocytosis of CD23 (Fcε receptor II) from the surface of B lymphocytes generates a 16-kDa intracellular fragment
AU Grenier-Brossette, Nicole; Bourget, Isabelle; Akoundi, Camelia; Bonnefoy, Jean Yves; Cousin, Jean Louis
CS Fac. Med. (Pasteur), Nice, F-06107, Fr.
SO European Journal of Immunology (1992), 22(6), 1573-7
CODEN: EJIMAF; ISSN: 0014-2980
DT Journal
LA English
AB It has been reported that the 45-kDa low-affinity **antibody** Fcε receptor (FcεRII) on B cells is cleaved spontaneously

from the cell surface to release soluble fragments. This study demonstrates an addnl. fate of the FcεRII. 125I- **labeled CD23** + B cells were cultured for 24 h at 37°. After lysis, cell exts. were immunoppt'd. with CD23 monoclonal **antibodies**. Using this methodol., it was demonstrated that an increasing amount of the labeled FcεRII becomes progressively resistant to externally applied trypsin, indicating that a fraction of the cell surface receptors are internalized. In parallel, a **labeled** 16-kDa material, recognized by **CD23** monoclonal **antibodies** directed to the lectin-like domain of the FcεRII appears inside the cells. **Chloroquine** does not affect internalization of the FcεRII, but completely abolishes the formation of the intracellular fragment, suggesting that the receptor is processed by proteolytic cleavage in acidic organelle. In addition, the internalization is enhanced in the presence of CD23 monoclonal **antibodies**. Thus, FcεRII can be internalized by ligand-induced endocytosis and subsequently cleaved in an intracellular compartment. These results also support the view that the FcεRII is involved in antigen focusing and antigen presentation.

L17 ANSWER 16 OF 24 CA COPYRIGHT 2004 ACS on STN DUPLICATE 10
AN 117:46201 CA
TI Internalization, lysosomal degradation and new synthesis of surface membrane CD4 in phorbol ester-activated T-lymphocytes and U-937 cells
AU Munck Petersen, C.; Ilsoe Christensen, E.; Storstein Andresen, B.; Moeller, B. K.
CS Dep. Clin. Immunol., Univ. Hosp. Aarhus, Den.
SO Experimental Cell Research (1992), 201(1), 160-73
CODEN: ECREAL; ISSN: 0014-4827
DT Journal
LA English
AB Protein kinase C activating phorbol esters downregulated membrane CD4 by endocytosis in U-937 and human T-cells. Half-time for internalization (.apprx.15 min at 50 ng/mL PMA) was determined by fluorescence-activated cell sorting. **CD4**-bound 125I-**labeled** anti-**CD4** **mAb** was rapidly degraded in PMA-activated cells, whereas degradation was low in resting cells. Endocytosis and/or degradation of anti-CD4 **mAb** was suppressed by H 7, and by inhibitors of membrane traffic (monensin) and lysosome function (**methylamine**, **chloroquine**). Immunocytochem. localized CD4 to the surface of unstimulated T-cells. Upon PMA stimulation occasional **labeling** was seen in endosomes but whole cell **CD4** decreased dramatically. However, **methylamine**-treated PMA blasts showed accumulation of CD4 in lysosomes and accordingly, pulse-chase expts. in biolabeled cell cultures suggested a manifest reduction of CD4 half-life in response to PMA. Despite their low surface CD4 d., PMA blasts exhibited uptake and accelerated degradation of anti-CD4 **mAb**. Also, inhibitors of protein synthesis enhanced the PMA-induced downregulation, and membrane CD4 reappeared on fully activated as well as unstimulated cells treated with trypsin. Ongoing **CD4** synthesis in activated cells was further evidenced by metabolic **labeling** and Northern blot anal. demonstrating unaltered or slightly increased **CD4** protein and mRNA levels resulting from PMA. Thus, phorbol esters downregulate the cellular CD4 pool by endocytosis and subsequent lysosomal degradation of membrane CD4. Transport of CD4 to the cell surface and CD4 synthesis is unaffected by activation.

L17 ANSWER 17 OF 24 MEDLINE on STN
AN 92042015 MEDLINE
DN PubMed ID: 1657957
TI A surface antigen of Giardia lamblia with a glycosylphosphatidylinositol anchor.
CM Erratum in: J Biol Chem 1991 Dec 5;266(34):23516

AU Das S; Traynor-Kaplan A; Reiner D S; Meng T C; Gillin F D
CS Department of Pathology, University of California, San Diego 92103.
NC AI 19863 (NIAID)
AI 24285 (NIAID)
AM 35108 (NIADDK)
+
SO Journal of biological chemistry, (1991 Nov 5) 266 (31) 21318-25.
Journal code: 2985121R. ISSN: 0021-9258.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199112
ED Entered STN: 19920124
Last Updated on STN: 19920124
Entered Medline: 19911213

NR

AB Since Giardia lamblia trophozoites are exposed to high concentrations of fatty acids in their human small intestinal milieu, we determined the pattern of incorporation of [3H]palmitic acid and myristic acid into G. lamblia proteins. The pattern of fatty acylation was unusually simple since greater than 90% of the Giardia protein biosynthetically labeled with either [3H]palmitate or myristate migrated at approximately 49 kDa (GP49) in reducing sodium dodecyl sulfate-polyacrylamide gel electrophoresis during both growth and differentiation. GP49, which partitions into the Triton X-114 detergent phase, is localized on the **cell surface** since it is 125I-surface-**labeled.**
GP49 was also biosynthetically labeled with [14C]ethanolamine and [3H]myoinositol, suggesting that it has a glycosylphosphatidylinositol (GPI) anchor. Moreover, phospholipase A2 (PLA2) or mild alkaline treatment released free fatty acids, indicating a diacylglycerol moiety with ester linkages. Finally, a 3H- and 14C-labeled species was released by nitrous acid deamination from [14C]palmitate- and [3H]myoinositol-labeled GP49. The GPI anchor of GP49 is unusual, however, because purified GP49 was cleaved by Bacillus cereus phosphatidylinositol (PI)-specific PLC, but not by Staphylococcus aureus PI-PLC, or plasma PLD, and did not react with **antibody** against the variant surface glycoprotein cross-reactive determinant. Moreover, the double-labeled deaminated GP49 anchor migrated faster than authentic PI in TLC and produced [3H]glycerophosphoinositol after deacylation. In contrast to the variable cysteine-rich G. lamblia surface antigens described previously, GP49 was identified in Western blots of every isolate tested, as well as in subclones of a single isolate which differ in expression of a major cysteine-rich 85/66-kDa surface antigen, which does not appear to be GPI-anchored. These observations suggest that GP49, the first common surface antigen to be described in G. lamblia, may play an important role in the interaction of this parasite with its environment.

no fluorescent label

L17 ANSWER 18 OF 24 CA COPYRIGHT 2004 ACS on STN DUPLICATE 11
AN 116:171922 CA
TI Mechanisms of serum-enhanced adhesion of human alveolar macrophages to epithelial cells
AU McGowan, Stephen E.; Heckman, Judith G.
CS Dep. Veterans Aff. Med. Cent., Iowa City, IA, 52242, USA
SO Lung (1991), 169(4), 215-26
CODEN: LUNGD9; ISSN: 0341-2040
DT Journal
LA English
AB Adhesive interactions between macrophages and epithelial cells in the pulmonary alveoli may be important in the pathogenesis of inflammatory lung diseases, such as those induced by cigarette smoking. Potential mechanisms controlling the interactions between these cells were investigated using human alveolar macrophages (AM) and MDCK or A549 epithelial cells. Five percent human serum enhanced the adhesion of AM to

MDCK cells by approx. 6-fold and to A549 cells by approx. 1.7-fold. This enhancement was reduced by heating the serum for 30 min at 55°. Treating normal human serum with **methylamine** to inactivate complement C3, substituting C3-deficient serum, or pretreating serum-exposed MDCK cells with anti-C3 F(ab')2 all significantly diminished the adhesion of AM, suggesting that complement is involved. With the use of **flow cytometry** to examine complement receptors on AM, both **CD11b/CD18** and **CD11c**/CD18 were detected but CR1 was not evident. Preincubating AM with a monoclonal **antibody** to CD18 reduced the adhesion of AM to MDCK cells by 40% while a significant reduction could not be demonstrated after preincubation with **antibodies** to either CD11b or CD11c. These data suggest that in the presence of serum C3bi is deposited on the surface of MDCK cells and that AM may attach to these cells, at least in part, through interactions between C3bi and one or more receptors in the CD11/CD18 family, which are present on AM.

L17 ANSWER 19 OF 24 CA COPYRIGHT 2004 ACS on STN
AN 115:251131 CA
TI Intracellular catabolism of radiolabeled anti-CD3 **antibodies** by leukemic T cells
AU Geissler, Francis; Anderson, Susan K.; Press, Oliver
CS Dep. Biol. Struct., Univ. Washington, Seattle, WA, 98195, USA
SO Cellular Immunology (1991), 137(1), 96-110
CODEN: CLIMB8; ISSN: 0008-8749
DT Journal
LA English
AB The endocytosis and intracellular metabolism of radiolabeled anti-CD3 **MoAb** 64.1 by the malignant human T cell line HPB-ALL were studied using biochem., morphol., electrophoretic, and chromatog. techniques. Biosynthetically labeled [3H]64.1 and externally radioiodinated 125I-64.1 were similarly internalized and degraded by tumor cells, with .apprx.70% of the initally bound radioactivity being released to the culture supernatant as TCA soluble radioactivity in the 1st 24 h of culture. Radiolabeled 64.1 was routed from the cell membrane to endosomes where initial proteolysis began and finally to lysosomes where terminal catabolism to single amino acids occurred. SDS-PAGE demonstrated 4 major intracellular metabolite species (46, 25, 15, and <10 kDa). Thin-layer chromatog. demonstrated that >95% of the TCA soluble radioactivity in culture supernatants was [125I]monoiodotyrosine, indicating that proteases, not deiodinases, were of primary importance in catabolism of 125I-64.1. In the presence of inhibitors of lysosomal function (leupeptin, monensin, and ammonium chloride), 125I-64.1 degradation was impeded, causing prolonged retention of radioactivity in the lysosomal compartment of cells. However, although the pace of catabolism was markedly diminished by these agents, no major changes in the sizes fo intermediate metabolites generated were observed The results suggest that judicious administration of lysosomal inhibitors (e.g., **chloroquine**, verapamil, monensin) may enhance retention of radioimmunoconjugates by lymphoid malignancies, improving radioimmunoscintigraphic and radioimmunotherapeutic efforts.

L17 ANSWER 20 OF 24 CA COPYRIGHT 2004 ACS on STN
AN 112:194518 CA
TI Inhibition of catabolism of radiolabeled **antibodies** by tumor cells using **lysosomotropic** amines and carboxylic ionophores
AU Press, Oliver W.; DeSantes, Kenneth; Anderson, Susan K.; Geissler, Francis
CS Dep. Med., Univ. Washington, Seattle, WA, 98195, USA
SO Cancer Research (1990), 50(4), 1243-50
CODEN: CNREA8; ISSN: 0008-5472
DT Journal
LA English
AB The rates of degradation of radioiodinated monoclonal **antibodies** (**MoAbs**) by malignant T- and B-lymphoid cells were studied in the

presence and absence of a variety of pharmacol. agents known to affect the intracellular metabolism of internalized ligands. 125I-**labeled MoAbs** directed against the **CD2**, **CD3**, **CD5**, and anti-μ surface antigens underwent rapid endocytosis, followed by prompt degradation with release of ≥50% of the initially bound radioactivity as free, TCA-soluble 125I within 24 h. **Lysosomotropic** amines (**chloroquine**, NH4Cl, amantadine), carboxylic ionophores (monensin, nigericin), Ca channel blockers (verapamil), thionamides (propylthiouracil), lysosomal enzyme inhibitors (leupeptin), and colchicine all inhibited metabolism of radioiodinated **MoAbs** and enhanced retention of 125I-labeled **MoAbs** by tumor cells. The most effective agents (e.g., monensin, nigericin) diminished the release of free 125I by >90% and enhanced retention of radioactivity by >300% at 24 h. Expts. with immunoperoxidase electron microscopy and Percoll gradient fractionation of organelles from disrupted cells suggested that high concns. of monensin (10-20 μM) delayed transfer of 125I-labeled **MoAbs** to lysosomes, but other mechanisms (e.g., pH neutralization) were operative at lower concns. (1-3 μM). Clin. administration of these agents may enhance retention of radioimmunoconjugates by tumor cells, resulting in improved radioimmunoscintigraphy and radioimmunotherapy.

L17 ANSWER 21 OF 24 CA COPYRIGHT 2004 ACS on STN DUPLICATE 12
AN 109:168582 CA
TI Monoclonal **antibody** internalization and degradation during modulation of the CD3/T-cell receptor complex
AU Schaffar, Laurence; Dallanegra, Anne; Breittmayer, Jean Philippe; Carrel, Stefan; Fehlmann, Max
CS Fac. Med., Nice, Fr.
SO Cellular Immunology (1988), 116(1), 52-9
CODEN: CLIMB8; ISSN: 0008-8749
DT Journal
LA English
AB Although it is well known that the CD3/T-cell receptor (TCR) complex modulates from the surface of T cells upon exposure to monoclonal **antibodies** (**mAb**) directed against it, the fate of bound **mAb** has not been yet elucidated. The authors therefore performed direct binding expts. of 125I-**labeled mAb** against **CD3** or TCR to investigate their fate in Jurkat T cells. All **mAb** were progressively internalized and degraded in Jurkat T cells and this degradation was inhibited by **chloroquine**, an inhibitor of lysosomal degradation enzymes. The sequestration of anti-CD3 **mAb** in acid compartments was further shown using cytofluorometry. Thus, **antibodies** against CD3 or against TCR follow the same endocytic pathway.

no fluorophore

L17 ANSWER 22 OF 24 MEDLINE on STN
AN 86168474 MEDLINE
DN PubMed ID: 3007532
TI Heterologous transmembrane and cytoplasmic domains direct functional chimeric influenza virus hemagglutinins into the endocytic pathway.
AU Roth M G; Doyle C; Sambrook J; Gething M J
NC AID 19630 (NIAID)
SO Journal of cell biology, (1986 Apr) 102 (4) 1271-83.
Journal code: 0375356. ISSN: 0021-9525.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 198605
ED Entered STN: 19900321
Last Updated on STN: 19970203
Entered Medline: 19860512

AB Chimeric genes were created by fusing DNA sequences encoding the ectodomain of the influenza virus hemagglutinin (HA) to DNA coding for the transmembrane and cytoplasmic domains of either the G glycoprotein of vesicular stomatitis virus or the gC glycoprotein of Herpes simplex virus 1. CV-1 cells infected with SV40 vectors carrying the recombinant genes expressed large amounts of the chimeric proteins, HAG or HAgC on their surfaces. Although the ectodomains of HAG and HAgC differed in their immunological properties from that of HA, the chimeras displayed the biological functions characteristic of the wild-type protein. Both HAG and HAgC bound erythrocytes as efficiently as HA did and, after brief exposure to an acidic environment, induced the fusion of erythrocyte and CV-1 cell membranes. However, the behavior of HAG and HAgC at the cell surface differed from that of HA in several important respects. HAG and HAgC were observed to collect in coated pits whereas wild-type HA was excluded from those structures. In the presence of **chloroquine**, which inhibits the exit of receptors from endosomes, HAG and HAgC accumulated in intracellular vesicles. By contrast, **chloroquine** had no effect on the location of wild-type HA. HAG and HAgC **labeled** at the **cell surface** exhibited a temperature-dependent acquisition of resistance to extracellular protease at a rate similar to the rates of internalization observed for many cell surface receptors. HA acquired resistance to protease at a rate at least 20-fold slower. We conclude that HAG and HAgC are efficiently routed into the endocytic pathway and HA is not. However, like HA, HAG was degraded slowly, raising the possibility that HAG recycles to the plasma membrane.

L17 ANSWER 23 OF 24 MEDLINE on STN
AN 85054964 MEDLINE
DN PubMed ID: 6094573
TI Complete inhibition of transferrin recycling by monensin in K562 cells.
AU Stein B S; Bensch K G; Sussman H H
NC CA09151 (NCI)
CA13533 (NCI)
SO Journal of biological chemistry, (1984 Dec 10) 259 (23) 14762-72.
Journal code: 2985121R. ISSN: 0021-9258.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 198501
ED Entered STN: 19900320
Last Updated on STN: 19970203
Entered Medline: 19850110
AB Monensin blocks human transferrin recycling in a dose-dependent and reversible manner in K562 cells, reaching 100% inhibition at a noncytocidal dose of 10(-5) M, whereas transferrin recycling is virtually unaffected by noncytocidal doses of **chloroquine**. The intracellular pathway of human transferrin in K562 cells, both in the presence and absence of 10(-5) M monensin, was localized by indirect immunofluorescence. Monensin blocks transferrin recycling by causing internalized ligand to accumulate in the perinuclear region of the cell. The effect of 10(-5) M monensin on human transferrin kinetics was quantitatively measured by radioimmunoassay and showed a positive correlation with immunofluorescent studies. Immunoelectron microscopic localization of human transferrin as it cycles through K562 cells reveals the appearance of perinuclear transferrin-positive multivesicular bodies within 3 min of internalization, with subsequent exocytic delivery of the ligand to the **cell surface** via transferrin-**staining** vesicles arising from these perinuclear structures within 5 min of internalization. Inhibition of ligand recycling with 10(-5) M monensin causes dilated transferrin-positive multivesicular bodies to accumulate within the cell with no evidence of recycling vesicles. A coordinated interaction between multivesicular bodies and the Golgi

apparatus appears to be involved in the recycling of transferrin in K562 cells. Cell-surface-binding sites for transferrin were reduced by 50% with 10(-5) M monensin treatment; however, this effect was not attenuated by 80% protein synthesis inhibition with cycloheximide, supporting the idea that the transferrin receptor is also recycled through the Golgi.

L17 ANSWER 24 OF 24 CA COPYRIGHT 2004 ACS on STN DUPLICATE 13
AN 98:69337 CA
TI Membrane proteins of the vacuolar system. III. Further studies on the composition and recycling of endocytic vacuole membrane in cultured macrophages
AU Muller, William A.; Steinman, Ralph M.; Cohn, Z. A.
CS Rockefeller Univ., New York, NY, 10021, USA
SO Journal of Cell Biology (1983), 96(1), 29-36
CODEN: JCLBA3; ISSN: 0021-9525
DT Journal
LA English
AB The membrane of macrophage phagolysosomes were selectively radioiodinated in living cells by phagocytosis of lactoperoxidase covalently coupled to latex spheres (LPO-latex), followed by iodination on ice with Na125I and H2O2. Three approaches were employed to examine the polypeptide composition of the phagolysosome (PL) and plasma membranes (PM). The efficiency of intracellular iodination was increased by increasing lysosomal pH with **chloroquine**. By 1-dimensional SDS polyacrylamide gel electrophoresis, the heavily labeled **chloroquine**-treated PL exhibited the same labeled polypeptides as PM iodinated extracellularly with LPO-latex. Iodinated PL and PM were compared by 2-dimensional gel electrophoresis. No differences in the isoelec. point and mol. weight of the major iodinated species were detected. Quant. immune precipitation was performed with 5 specific **antibodies** directed against cell surface antigens. Four **antibodies** precipitated similar relative amts. of **labeled** antigen on the **cell surface** and endocytic vacuole. One **antibody**, secreted by hybridoma 2.6, detected a 21-kilodalton polypeptide that was enriched 7-fold in PL membrane. This enrichment was **cell surface** derived, since the amount of **labeled** 2.6 was increased 7-fold when iodinated PM was driven into the cell during latex uptake. Therefore, intracellular iodination primarily detects PL proteins that are identical to their PM counterparts. Addnl. studies employed electron microscope autoradiog. to monitor the centrifugal flow of radiolabeled polypeptides from PL to PM. Cells were iodinated intralysosomally and returned to culture for only 5-10 min at 37°. Most of the cell-associated **label** then redistributed to the **cell surface** or its adjacent area. Significant movement out of the lysosome compartment occurred even at 2° and 22°. Extensive and rapid membrane flow through the secondary lysosome presumably contributes to the great similarity between PM and PL membrane polypeptides.

=> log y

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1	894	(435/7.24).CCLS.	USPAT; US-PGPUB; EPO; DERWENT	2004/07/06 15:05
2	735	(435/961-962).CCLS.	USPAT; US-PGPUB; EPO; DERWENT	2004/07/06 15:05
3	1337	(435/975).CCLS.	USPAT; US-PGPUB; EPO; DERWENT	2004/07/06 15:05
4	733	(436/800).CCLS.	USPAT; US-PGPUB; EPO; DERWENT	2004/07/06 15:06
5	840	(436/808).CCLS.	USPAT; US-PGPUB; EPO; DERWENT	2004/07/06 15:06
6	314	(436/826).CCLS.	USPAT; US-PGPUB; EPO; DERWENT	2004/07/06 15:07
7	4015	((435/7.24).CCLS.) or ((435/961-962).CCLS.) or ((435/975).CCLS.) or ((436/800).CCLS.) or ((436/808).CCLS.) or ((436/826).CCLS.)	USPAT; US-PGPUB; EPO; DERWENT	2004/07/06 15:07
8	341	lysosomotropic	USPAT; US-PGPUB; EPO; DERWENT	2004/07/06 15:07
9	3649	chloroquine	USPAT; US-PGPUB; EPO; DERWENT	2004/07/06 15:08
10	864	hydroxychloroquine	USPAT; US-PGPUB; EPO; DERWENT	2004/07/06 15:09
11	664	primaquine	USPAT; US-PGPUB; EPO; DERWENT	2004/07/06 15:09
12	29835	methylamine	USPAT; US-PGPUB; EPO; DERWENT	2004/07/06 15:09
13	34013	lysosomotropic or chloroquine or hydroxychloroquine or primaquine or methylamine	USPAT; US-PGPUB; EPO; DERWENT	2004/07/06 15:10
14	180352	antibody or moab or mab or mcab	USPAT; US-PGPUB; EPO; DERWENT	2004/07/06 15:11
15	636	(lysosomotropic or chloroquine or hydroxychloroquine or primaquine or methylamine) same (antibody or moab or mab or mcab)	USPAT; US-PGPUB; EPO; DERWENT	2004/07/06 15:11
16	13	(((435/7.24).CCLS.) or ((435/961-962).CCLS.) or ((435/975).CCLS.) or ((436/800).CCLS.) or ((436/808).CCLS.) or ((436/826).CCLS.)) and ((lysosomotropic or chloroquine or hydroxychloroquine or primaquine or methylamine) same (antibody or moab or mab or mcab))	USPAT; US-PGPUB; EPO; DERWENT	2004/07/06 15:11